DESCRIPTION OF THE FIGURES

FIG. 1 provides a schematic representation of a fibronectin molecule, including chymotryptic fragments.

FIG. 2 shows the infection efficiency of committed human progenitor cells in the presence of fibronectin fragments using the TKNEO vector, as further described in Example 1, *infra*.

FIG. 3 compares the infection efficiency of various committed human hematopoietic progenitor cells in the presence of fibronectin fragments thereof using the TKNEO vector, as further described in Example 1, *infra*.

FIG. 4 compares the presence of hADA in mice engrafted with bone marrow cells transduced by (i) the coculture method (lanes 2-4), (ii) supernatant infection in the presence of immobilized fibronectin fragments (lanes 5-7), and supernatant infection on BSA (lanes 8-10), as further described in Example 7, *infra*. Controls for hADA are shown in lanes 1 and 12 and for murine ADA in lane 11.

FIG. 5 demonstrates retroviral binding to fibronectin fragments, as further described in Example 8, *infra*.

FIG. 6 demonstrates that retroviral binding to fibronectin fragments is dose-dependent, as further described in Example 8, *infra*.

FIG. 7 provides a schematic diagram illustrating various recombinant fibronectin fragments used in Examples 9-11, *infra*.

FIG. 8 shows retrovirus binding to various fibronectin fragments, including to several recombinant fragments, as described in Example 9, *infra*.

FIG. 9 demonstrates that heparin blocks retrovirus binding to fibronectin fragments, as described in Example 9, *infra*.

FIG. 10 shows the efficiency of retrovirus infection of murine hematopoietic cells in the presence of various fibronectin fragments, as further reported in Example 10, *infra*.

FIG. 11 compares the presence of hADA in mice engrafted with bone marrow cells transduced by (i) the coculture method, (ii) supernatant infection on various fibronectin fragments, and (iii) supernatant infection on BSA, as described in Example 11, *infra*.

DESCRIPTION OF THE PREFERRED EMBODIMENTS

For the purpose of promoting an understanding of the principles of the invention, reference will now be made to certain embodiments thereof and specific language will be used to describe the same. It will nevertheless be understood that no limitation of the scope of the invention is thereby intended, such alterations, further modifications and such applications of the principles of the invention as illustrated herein being contemplated as would normally occur to one skilled in the art to which the invention relates.

As indicated above, the present invention provides methods for increasing the frequency of transduction of viable cells by viruses such as retroviruses. The invention also provides methods for efficient gene transfer into viable cells using recombinant retroviral vectors, methods for obtaining transduced cells, and methods and materials for achieving autologous and other cellular grafts.

One feature of the present invention is the discovery that fibronectin (FN), and fragments of fibronectin containing the CS-1 cell-adhesion domain of FN, significantly enhance retroviral-mediated gene transfer into cells such as hematopoietic cells, e.g. committed progenitors and primitive hematopoietic stem cells or long-term culture-initiating cells (LTC-IC), carrying a fibronectin receptor and thereby exhibiting the capacity to bind to fibronectin

or fragments thereof. Advantageously, this increased efficiency makes cocultivation with virus-producing cells unnecessary. Other features of the invention capitalize on the discovery of a viral-binding domain of fibronectin located within the Heparin-II binding domain. This viral-binding domain can be used to localize virus particles in many applications, including for example in a broad range of constructs for delivering the virus to a target cell.

Recombinant viral vectors in accordance with certain preferred aspects of the present invention contain exogenous DNA and are non-pathogenic, i.e. replication-defective. These vectors efficiently transfer and precisely and stably integrate exogenous DNA into cellular DNA of host cells such as animal cells, particularly mammalian cells. For example, in the present invention a nucleotide sequence including a run of bases from the coding sequence of the gene of interest can be incorporated into a recombinant retroviral vector under the control of a suitable promoter to drive the gene, typically an exogenous promoter. In this regard, the exogenous DNA can contain DNA which has either been naturally or artificially produced, and can be from parts derived from heterologous sources, which parts may be naturally occurring or chemically synthesized molecules, and wherein those parts have been joined by ligation or other means known to the art. As indicated, the introduced nucleotide sequence will be under control of a promoter and thus will be generally downstream from the promoter. Stated alternatively, the promoter sequence will be generally upstream

(i.e., at the 5' end) of the coding sequence. In this vein, it is well known that there may or may not be other regulatory elements (e.g., enhancer sequences) which cooperate with the promoter and a transcriptional start codon to achieve transcription of the exogenous coding sequence. The phrase "under control of" contemplates the presence of such other elements as are necessary to achieve transcription of the introduced gene. Also, the recombinant DNA will preferably include a termination sequence downstream from the introduced coding sequence.

Retroviral vectors that include exogenous DNA providing a selectable marker or other selectable advantage can be used. For example, the vectors can contain one or more exogenous genes that provide resistance to various selection agents including antibiotics such as neomycin. Representative vectors which can be used in the invention include for example the N_2/ZipTKNEO vector (TKNEO) (titer: $1x10^5$ $G418^r$ cfu/ml on NIH 3T3 cells), the ZipPGK-hADA vector, and the ZipPGK-mADA vector all as previously reported by Moritz et al. (1993) *J. Exp. Med.* 178:529. In the TKNEO vector, neo phosphotransferase sequences are expressed in the sense orientation (relative to the 5' long terminal repeat-LTR) via the herpes simplex thymidine kinase promoter. This vector contains a selectable marker gene which provides neomycin resistance to facilitate the identification of transduced cells. In the ZipPGK-hADA vector, the human ADA ("hADA") cDNA is expressed in the sense orientation relative to the 5'LTR via the human phosphoglycerate